

## **Influence of the sample preparation (dehydration) on the plasmatic membrane cell morphology: HeLa cancer cells visualized by AFM.**

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The neoplastic transformation in the membrane morphology is an important indicator of the cancer progress. Recent involvement of the nanotechnology into cancer treatments via uptake of the medicine attached to the nanoparticles, controlled by the membrane porosity, in addition, intensify our interest into characterization of the cell surface.

Here, the AFM was used to characterize the surface properties of the HeLa cellular line, isolated from cervical cancer. The cellular material was cultivated directly on the gold film substrates and before visualization samples were rinsed (dehydrated) by the ethanol-water solution, with different content of ethanol: Group-A [70-100%] and Group-B [20,-100%], in order to see the influence of dehydration on the cell morphology and the size of the membrane pores. The AFM images obtained by Nanoscope IIIa AFM Multimode, in the tapping mode, revealed detail morphology of the HeLa plasmatic membrane on the nanometric-molecular level. In both group a characteristic star-shaped cells, were observed. In Group-A, on the external membrane surface homogenous pores of 395 nm of diameter (circular shape), were found. In the Group-B, with less intensity of dehydration, the membrane surface posses rather oval shape pores with dimensions of 306 nm and 446 nm, which is closer to the some literature data. In addition, the interaction force measurements between the AFM tip and HeLa membrane (attraction and adhesion) were evaluated and compared to give us more information about the membrane mechanical properties. Analyses of these data are in the progress.

As an achievement in our study, we like to emphasize: development of new sample preparation procedure for growing cells on the flat gold film, imaging of the plasmatic membrane surface at the high resolution to reveled morphology on the nanometric level, not previously achieved, by another type of microscopes.

- Camesano T.A., Natan M.J., Logan B.E., 2000, Observation of Changes in Bacterial Cell Morphology Using Tapping Mode Atomic Force Microscopy, Langmuir, 16:4563-4572.
- Simon A., Cohen-Bouhacina T., Porté M.C., Aimé J.P., Amédée J., Bareille R. y Baquery C., 2003, Characterization of Dynamic Cellular Adhesion of Osteoblasts Using Atomic Force Microscopy, Wiley-Liss Int., 54: 36-47.
- Velegol S.B., Pardi S., Li X., Velegol D. y Logan B.E., 2003, AFM Imaging Artifacts due to Bacterial Cell Height and AFM tip Geopmetry, Langmuir, 19:851-857.

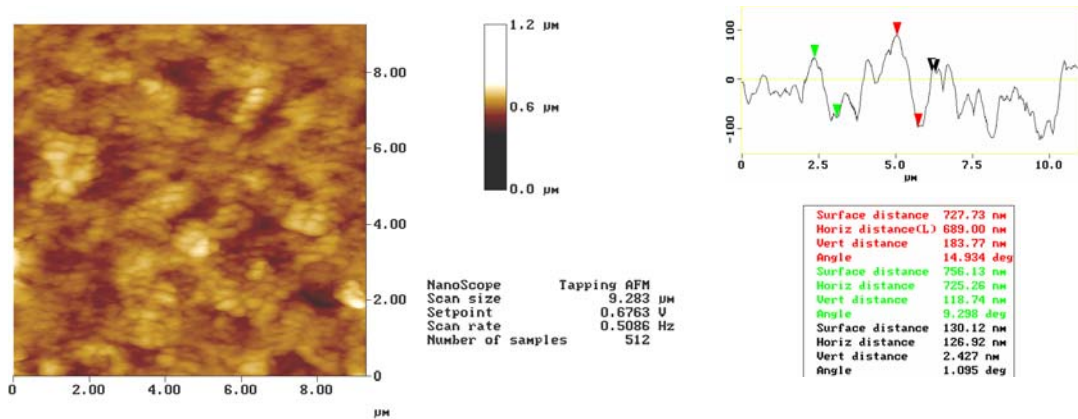


Figure 1. Image of the surface of the plasmatic mebran of the cellular line HeLa. It was dehydrated in a series of ethanol-water from 70% to 100%. The line of topographical profile presents the present reliefs in the surface and the marks it indicates us the exact value of the heights and longitudes of these structures. AFM / Tapping mode (9.28μm x 9.28μm).

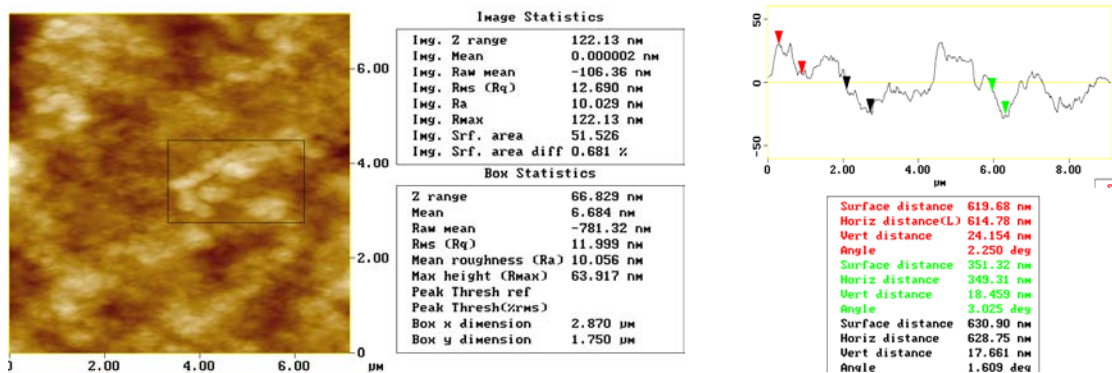


Figure 2. Image of the surface of the plasmatic mebran of the cellular line HeLa. It was dehydrated in to series of ethanol-water from 70% to 100%. The line of topographical and the analysis of ruggedness of the surface, allow us to know the existent variations in the topography of the surface of plasmatic membrane and to be able to study the impact from the methodologies of dehydration to ultrastructural level. (6.68μm x 6.68μm).